Society for Endocrinology
European Society of Endocrinology

*Endocrine Connections* (2025) **14** e240437
https://doi.org/10.1530/EC-24-0437

Received 18 August 2024
Accepted 23 December 2024
Available online 24 December 2024
Version of Record published 17 January 2025

RESEARCH

# Magnetic resonance imaging does not distinguish Kallmann syndrome from normosmic isolated hypogonadotropic hypogonadism

Małgorzata Kałużna[1,2], Katarzyna Katulska[3], Katarzyna Ziemnicka[1,2], Pola Kompf[1], Bartłomiej Budny[1,2], Paweł Komarnicki[1,2], Michał Rabijewski[4], Jerzy Moczko[5], Jarosław Kałużny[6] and Marek Ruchała[1,2]

[1]Department of Endocrinology, Metabolism and Internal Medicine, Poznań University of Medical Sciences, Poland
[2]Ward of Endocrinology, Metabolism and Internal Diseases Ward, University Clinical Hospital, Poznan, Poland
[3]Department of General Radiology and Neuroradiology, General Radiology Unit, Poznań University of Medical Sciences, Poland
[4]Department of Reproductive Health, Centre of Postgraduate Medical Education, Warsaw, Poland
[5]Department of Computer Science and Statistics, Poznań University of Medical Sciences, Poland
[6]Department of Otolaryngology, Head and Neck Surgery, Poznań University of Medical Sciences, Poland

Correspondence should be addressed to M Kałużna: mkaluzna@ump.edu.pl

## Abstract

*Introduction and objectives:* Isolated hypogonadotropic hypogonadism (IHH) may be associated with pituitary gland and olfactory system disorders. We aimed to correlate findings of magnetic resonance imaging (MRI) of the pituitary gland and olfactory system in IHH patients with the patients' olfactory phenotype.

*Patients and methods:* The present research was a single-center retrospective case–control study. MRI patterns of the pituitary gland and olfactory system were studied in 46 patients, of whom 29 (63%) were classified on the basis of olfactometry as having Kallmann syndrome (KS) (16 patients with anosmia and 13 patients with hyposmia) and 17 (37%) as having normosmic IHH (nIHH). Results were compared with age- and sex-matched healthy controls. Genetic diagnosis was conducted in all IHH patients based on next-generation sequencing.

*Results:* Almost 70% prevalence of pituitary hypoplasia was observed in IHH subjects. Olfactory bulb (OB) abnormalities were identified in 80.4% of all patients, both the KS (82.8%) and the nIHH (76.5%) subjects. Incidence of unilaterally abnormal, hypoplastic olfactory sulcus (OS) was equally frequent in nIHH and KS. Statistically, piriform cortical thickness was significantly lower in all patient groups than in controls.

*Conclusions:* MRI cannot exclusively differentiate between KS and nIHH, as both conditions may present with OB and OS abnormalities. A surprisingly high frequency of olfactory system abnormalities was observed in nIHH patients, while anterior pituitary hypoplasia was prevalent across all IHH patients. Notably, OB abnormalities were more predominant in KS patients than in those with nIHH.

Keywords: isolated hypogonadotropic hypogonadism (IHH); kallmann syndrome (KS); olfactory bulbs; olfactory sulcus; pituitary hypoplasia

Published by Bioscientifica Ltd.
https://ec.bioscientifica.com

## Introduction

Isolated hypogonadotropic hypogonadism (IHH), also known as isolated gonadotropin-releasing hormone (GnRH) deficiency, is defined by abnormally low serum levels of gonadotropins – luteinizing hormone (LH) and follicle-stimulating hormone (FSH) – accompanied by reduced circulating concentrations of sex steroids. This condition arises from insufficient secretion of hypothalamic GnRH. IHH can present with a normal sense of smell (normosmic IHH (nIHH)), observed in approximately 40% of cases, or with impaired olfactory function – ranging from partial (hyposmia) to complete loss (anosmia) – which characterizes Kallmann syndrome (KS), accounting for about 60% of IHH cases. IHH primarily affects sexual development, while in KS, olfactory development is also disrupted (1, 2). IHH is a rare but significant cause of pubertal disorders and infertility. In males, congenital IHH has a reported incidence of 1:10,000, while KS is estimated between 1:30,000 and 1:84,000. IHH in females is even less common, with male-to-female ratio of up to 4:1 in nIHH and up to 8:1 in KS (3, 4, 5).

Magnetic resonance imaging (MRI) is effective in visualizing pituitary dimensions and morphological anomalies of the pituitary and olfactory system (6). Hypothalamus and pituitary region MRI is advised to accurately diagnose IHH, and olfactory bulb (OB) MRI is recommended in patients with suspicion of dysosmia (7). Pituitary and olfactory system MRI in IHH enables non-invasive analysis of subtle morphological abnormalities in these areas. High-resolution fast spin-echo T2-weighted and T1-weighted imaging of the olfactory apparatus is preferred in the diagnostics of KS (8, 9). Olfactory defects identified using MRI may warrant examinations toward congenital anomalies. MRI could be useful for IHH diagnosis in pre-pubertal patients with anosmia or hyposmia. However, despite its diagnostic potential, MRI seems to be underutilized in IHH.

IHH has been associated with pituitary disorders, including anterior pituitary hypoplasia (APH), empty sella syndrome and microadenoma (10). APH was found in 6 of 16 patients with KS in one study (11). Dallago and coworkers described two cases of KS and empty sella syndrome (12). Non-functional pituitary microadenoma is a rare finding associated with IHH (10, 13). Coexistence of KS and craniopharyngioma was described by Jonklaas and coworkers (14). Pituitary MRI appears indispensable in IHH diagnostics.

### Aims

We aimed to measure the size and volume of the pituitary gland and OBs, olfactory sulcus depth and length and olfactory region cortical thickness using MRI in patients diagnosed with IHH, both nIHH and KS, and to correlate obtained MRI findings with patients' olfactory phenotype.

## Patients and methods

### Patient characteristics

This was a single-center retrospective case–control study. The basic inclusion criteria included delayed puberty and/or clinical symptoms of hypogonadism and low levels of LH, FSH and testosterone or estradiol (in women). Subjects with other endocrine disorders and acquired hypogonadism causes (e.g., hyperprolactinemia, pituitary macroadenoma, severe weight loss, drug-induced hypogonadism and excessive physical activity) were excluded.

IHH was diagnosed based on the following criteria: i) manifestation of absent, delayed or partial puberty or adult-onset isolated IHH (AO-IHH); ii) presence of clinical indications or symptoms suggestive of hypogonadism; iii) observance of low or normal serum concentrations of LH and FSH, typically below 4 IU/L, accompanied by a serum testosterone level less than 3.5 nmol/L in males or a serum estradiol level less than 20 pg/mL in females; iv) confirmation of otherwise normal anterior pituitary function; v) absence of prior surgical interventions within the sellar region; vi) maintenance of a normal serum ferritin concentration within the range (15, 16, 17); and vii) nIHH diagnosed in case of documentation of a normal sense of smell, both reported and confirmed (15, 16, 17).

AO-IHH was diagnosed based on an isolated failure of gonadotropin secretion that developed after otherwise normal sexual maturation or partial/complete pubertal development during adolescence. This diagnosis was further supported by evidence of the hypothalamic–pituitary–gonadal axis dysfunction in adulthood, as outlined by the criteria described above (18). Reversal of idiopathic hypogonadotropic hypogonadism was defined as the ability to maintain a normal adult endogenous serum testosterone concentration (≥9.4 nmol/L) after discontinuing hormonal therapy. The required duration of discontinuation was at least 2 weeks for pulsatile GnRH therapy, 4 weeks for transdermal testosterone and 6–8 weeks for injectable testosterone or human chorionic gonadotropin (18).

Forty-six patients diagnosed with IHH, including 41 males and 5 females, aged 18–57 years (mean (SD) = 29.2 (8.4) years; median (interquartile range (IQR)) = 28 (23–33)), underwent MRI of the hypothalamic–pituitary axis and olfactory region between 2013 and 2019. Twenty-four male and three female healthy controls, aged 18–48 years (mean (SD) = 29 (9.7) years; median (IQR) = 28 (18–37)), were also enrolled. Neither age nor sex distribution was significantly different between groups. All IHH patients had a well-established

reproductive and non-reproductive phenotype. Patients with other causes of hypothalamic–pituitary dysfunction (e.g., septo-optic dysplasia) were excluded.

Each IHH patient underwent a detailed medical interview and physical examination, including anthropometric measurements. Andrological assessment of male patients involved physical examination and testicular ultrasound. Female patients underwent a gynecological examination, with vaginal or transrectal ultrasound. Abdominal ultrasound was performed in all cases to identify potential congenital anomalies.

Serum pituitary hormones and corresponding target gland hormone levels were measured with a Cobas 6000 analyzer (Roche Diagnostics, Switzerland) and dedicated electrochemiluminescence immunoassays.

Detailed characteristics of the study subjects are shown in Table 1.

## Genetic testing

We employed next-generation sequencing (NGS) technology alongside a gene panel previously documented and reported for patient cohorts with KS and nIHH (19, 20). In particular, the Ion Torrent Personal Genome Machine System (Ion PGM TM, Thermo Fisher Scientific, Inc., USA) was utilized, and a validated panel comprising 38 genes associated with IHH and congenital hypopituitarism was applied. The gene panel included the following genes: *ADAM7*, *ANOS1*, *BMP2*, *BMP4*, *CHD7*, *FGF8*, *FGF17*, *FGFR1*, *GLI2*, *GNRH1*, *GNRHR*, *HESX1*, *HS6ST1*, *IGSF10*, *KISS1*, *KISS1R*, *LEP*, *LEPR*, *LHB*, *LHX3*, *LHX4*, *LRRIQ3*, *NSMF*, *NR0B1*, *OTX1*, *OTX2*, *PCSK1*, *PITX1*, *PITX2*, *PROK2*, *PROKR2*, *PROP1*, *POU1F1*, *SEMA3A*, *SOX3*, *TAC3*, *TACR3* and *WDR11*. Complete methodology for conducting genetic research, including the databases and software used for bioinformatics analysis, is available in our previous publications (19, 21).

## Olfactory testing

Regardless of the reported sense of smell, the olfactory function of each subject was tested according to the Elsberg–Levy olfactometry (ELO) with modifications by Pruszewicz, measuring olfactory thresholds using four different odorants (22, 23, 24). ELO is one of the most popular, available and useful olfactory assessment methods in Poland (25). Subsequently, patients were divided into anosmic, hyposmic and normosmic groups. The research was conducted between 2015 and 2018, before COVID-19 pandemic.

## MRI protocol

All pituitary examinations were performed in a 1.5T MR system (Magnetom Aera, Siemens Medical Systems, Germany) using a standard head coil (20 channels). T1-weighted spin-echo images were obtained at 550 ms/17 ms (TR). T2-weighted fast spin-echo images were obtained at 4,000 ms/95 ms (TR). For both sequences, the matrix used was 230 × 256 and the FOV was 170 × 170 mm.

Pituitary height was determined from midline sagittal images by measuring the greatest distance between the superior and inferior borders of the gland.

**Table 1** Baseline characteristics of patients with isolated hypogonadotropic hypogonadism.

| Parameter | All | nIHH | KS |
|---|---|---|---|
| N | 46 | 17 | 29 |
| Gender M/F (*n*; %) | 42/4; 91.3/9.7 | 17/0; 100/0 | 25/4; 86.2/13.8 |
| Age | 29.2 (9.7) | 28.94 (8.16) | 29.39 (8.63) |
| LH (mIU/mL) | 1.6 (1.7) | 1.9 (1.4) | 1.5 (2.0) |
| FSH (mIU/mL) | 1.7 (1.8) | 2.2 (1.7) | 1.4 (1.8) |
| T* (nmol/L) | 5.70 (4.19) | 5.10 (4.04) | 6.16 (4.37) |
| E2** (pg/mL) | 18.32 (19.41) | 14.06 (10.89) | 20.29 (22.19) |
| SHBG (nmol/L) | 42.60 (32.23) | 45.75 (39.70) | 40.59 (27.13) |
| TSH (μU/mL) | 2.10 (0.94) | 2.02 (0.91) | 2.15 (0.98) |
| FT4 (pmol/L) | 16.55 (3.49) | 16.79 (2.87) | 16.41 (3.87) |
| ACTH (pg/mL) | 38.22 (25.58) | 42.70 (36.87) | 35.64 (16.32) |
| Cortisol 8 AM (nmol/L) | 395.33 (141.86) | 381.18 (169.70) | 403.93 (124.62) |
| Cortisol 6 PM (nmol/L) | 162.69 (81.32) | 148.21 (82.90) | 170.80 (80.98) |
| IGF-1 (ng/mL) | 246.03 (121.14) | 249.0 (203.94) | 244.00 (138.59) |
| DHEAS (μg/dL) | 287.97 (166.76) | 264.48 (174.86) | 301.39 (163.68) |
| Max. LH† (mIU/mL) | 10.38 (8.25) | 12.85 (9.89) | 8.81 (6.86) |
| Max. FSH† (mIU/mL) | 3.71 (2.62) | 3.92 (2.26) | 3.58 (2.88) |

Data are presented as mean (SD); * in males; ** in females; † in a test with 100 μg intravenous gonadoliberin (LHRH); ACTH, adrenocorticotropic hormone; DHEAS, dehydroepiandrosterone sulfate; E2, estradiol (measured in women); FSH, follicle-stimulating hormone; FT4, free thyroxine; FTI, free testosterone index; IGF-1, insulin-like growth factor 1; KS, Kallmann syndrome; LH, luteinizing hormone; nIHH, normosmic isolated hypogonadotropic hypogonadism; SHBG, sex hormone-binding globulin; T, testosterone total (measured in men); and TSH, thyroid-stimulating hormone.

*Endocrine Connections* (2025) **14** e240437
https://doi.org/10.1530/EC-24-0437

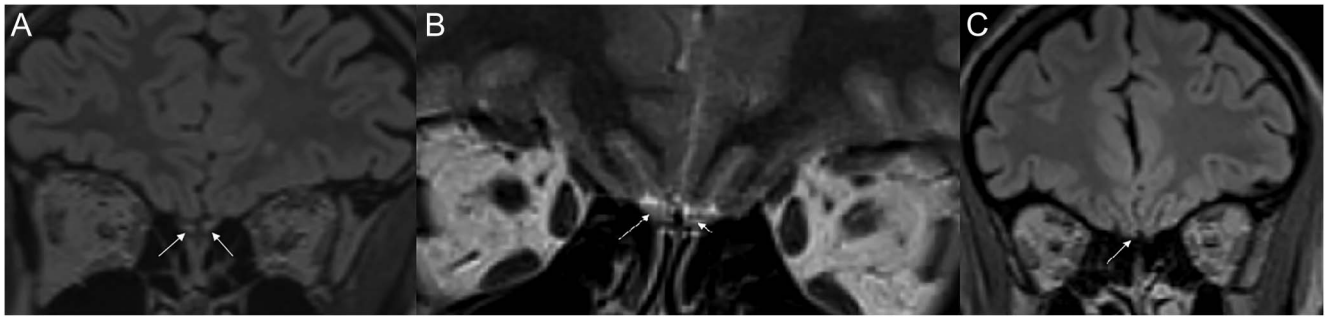


**Figure 1**

Coronal T2-weighted MRI scans from (A) a control subject with normal OBs; (B) a hyposmic IHH patient with asymmetric OBs – on the left side, OB not visible (short arrow) and on the right side, OB visible (long arrow); and (C) an anosmic IHH patient – bilateral absence of OB structures (the area of missing OBs is marked with a white arrow).

Lateral and anteroposterior dimensions were determined by measuring the greatest dimensions on the coronal and sagittal images. Pituitary volume was estimated using the following formula: $V$ = length × width × height × 0.52 (the last parameter was calculated with following formula: $((4/3)\pi r3)/a3$). Pituitary hypoplasia was diagnosed when the pituitary height was less than 5 mm (26). The pituitary gland was considered hypoplastic if its volume was reduced, i.e., it was lower than control group's mean pituitary volume minus 2 × SD.

All OB and olfactory sulcus (OS) examinations were performed in a 1.5T MR system (Magnetom Aera, Siemens Medical Systems, Germany) using a standard head coil (20 channels). Coronal turbo spin-echo T2-weighted images of all patients and the control group were obtained using the same parameters: FOV = 240 mm, slice thickness = 3.0 mm, repetition time (TR) = 5,400 ms, TE = 99 ms, matrix 512 × 512 and no space between slices or interpolation. The volumetric 3D magnetization-prepared rapid acquisition of gradient echo (MPRAGE) sequences, T1-weighted, was performed with the following parameters: axial acquisition, field-of-view (FOV) = 255 mm, TR = 9.7 ms, TE = 4.0 ms, flip angle = 12°, matrix = 512 × 224 and 1 $mm^3$ isometric voxel. Multi-planar reformation was conducted in sagittal and coronal planes.

OB volumes in KS patients and control subjects were measured, where the bulbs were visible, using coronal T2 images (Fig. 1). OBs and OSs were assessed in MRI according to Rombaux method (27). The bulb area was measured in consecutive slices and multiplied by the slice thickness to obtain its volume (27). Aplasia was recorded in cases where no bulb was identified. T2 images were used to assess the maximum OS depth. Its length was measured in the axial reformation of 3D T1 datasets, parallel to the cribriform plate (27).

OB was considered hypoplastic if its volume was reduced, i.e., it was lower than control group's mean bulb volume minus 2 × SD. The OS was recognized as hypoplastic if it was reduced, with its length or depth lower than mean minus 2 × SD of the control group's respective measurement (27, 28)

MRI measurements of piriform cortical thickness were also made (Fig. 2) (29). All measurements were made by an experienced radiologist to whom olfactory test results had not been disclosed. MRI descriptions were made by a radiologist trained in the assessment of this type of MRI, followed by two further verifications.

## Statistical analysis

Descriptive statistics for quantitative variables were expressed as mean and SD. A Shapiro–Wilk test was used to determine normal distribution of data. Comparisons between groups were performed by the Mann–Whitney test and Pearson's chi-squared test. Statistical significance was set at $P < 0.05$, and analyses were conducted using the Statistica v.13.0 statistical

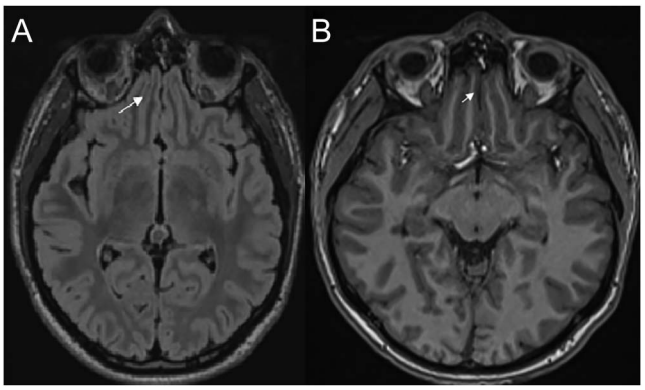


**Figure 2**

(A) T2-weighted MRI scan of olfactory cortex from a control subject (diameter 5 mm); (B) T1-weighted MRI scan of olfactory cortex from an anosmic IHH patient (diameter 3 mm).

**Table 2** Clinical features and MRI measurements in individual patients.

| | nIHH | KS | Controls |
|---|---|---|---|
| Sex | 17M, 1F | 25M, 4F | 24M, 3F |
| Age at MRI | 28.9 (8) | 29.4 (9) | 29 (10) |
| Olfaction | 17N | 13H, 16A | N |
| Right OBV ($mm^3$) | 50.1 (18) | 31.5 (26) | 72.6 (9) |
| Right OS length (mm) | 20.3 (4) | 17.2 (5) | 36.9 (5) |
| Right OS depth (mm) | 10.5 (2) | 8.7 (3) | 15.5 (1) |
| Left OBV ($mm^3$) | 42.6 (17) | 25.4 (28) | 72.4 (8) |
| Left OS length (mm) | 18.9 (3) | 16.6 (4) | 36.5 (5) |
| Left OS depth (mm) | 9.4 (2) | 7.9 (3) | 15.9 (1) |
| Olfactory cortex thickness (mm) | 3.9 (1) | 3.6 (1) | 4.7 (1) |
| Pineal cyst | 3 | 6 | 0/27 |
| PGV ($mm^3$) | 243 (110) | 220 (129) | 395.7 (70) |

A, anosmia; H, hyposmia; KS, Kallmann syndrome; N, normosmia; nIHH, normosmic isolated hypogonadotropic hypogonadism; OBV, olfactory bulb volume; OS, olfactory sulcus; and PGV, pituitary gland volume. All values in the table besides sex, olfaction and pineal cyst are presented as mean (SD).

software. To evaluate the agreement between the ELO and MRI quantitative analysis, Cohen's kappa inter-rater reliability index was used (MedCalc v.19.0.5).

# Results

## IHH patients' characteristics

Thirteen (28%) patients were diagnosed with adult-onset IHH. A reversal form of IHH was identified in 2 (4%) patients. Congenital defects were observed in 11 (24%) patients. Two patients presented with syndactyly affecting the second and third toes. Other isolated anomalies included aortic coarctation, arachnodactyly, ectopic posterior pituitary, oligophrenia, obturator hernia, cleft lip and palate, agenesis of the left kidney, splenomegaly, right nostril atresia, clinodactyly, a posterior arch defect of L5, incomplete kidney rotation and an abdominal hernia. In addition, 5 (11%) were diagnosed with primary hypothyroidism. Table 2 and Supplementary Table S1 (see section on Supplementary materials given at the end of the article) show the clinical characteristics of IHH participants, including qualitative and quantitative olfactory system MRI features, compared with the control group average values. Tables 3 and 4 present the NGS results. Pathogenic/likely pathogenic (P/LP) defects were identified in 15 (32.6%) patients, variants of uncertain significance (VUS) were identified in 22 (47.8%) patients, benign variants were identified in 3 (6.5%) patients, and no defects were found in 6 patients.

## Olfactory testing

Seventeen (36.9%) IHH patients were diagnosed as normosmic, 16 (34.8%) as anosmic, and 13 (28.3%) as hyposmic. All controls were reported as normosmic and were not referred to ELO.

## MRI results in control group

The control subjects had symmetrical, normal OBs (mean (SD) volume – right: 72.6 (9) $mm^3$; left: 72.4 (8) $mm^3$, $P > 0.05$) with fully visible olfactory tracts (Fig. 1A). OS lengths and depths were normal (mean (SD) lengths: right = 36.9 (5) mm, left = 36.5 (5) mm; depths: right = 15.5 (1) mm, left = 16 (1) mm). All control subjects had normal anterior pituitary height and volume. No pituitary region pathologies were found in controls (Table 5).

## MRI results in IHH patients

### Olfactory bulbs

Bilateral agenesis of the OBs in the brain MRI accompanied by anosmia in the ELO test was detected in 5 KS patients. Unilateral agenesis of the OB was diagnosed in 10 patients (2 anosmic and 8 hyposmic) (Fig. 1B). In all patients with unilateral bulb aplasia, their contralateral bulbs were identified as hypoplastic. Bilaterally hypoplastic OBs were observed in 27 IHH patients – in 11/16 (68.8%) of anosmic subjects, in 9/13 (69.2%) of hyposmic subjects and in 7/17 (41.2%) of nIHH subjects (Fig. 1C, Tables 2, 5). Table 5 presents the characteristics of olfactory structures and the pituitary gland observed in the MRI of IHH participants, divided into three olfactory categories.

### Olfactory tracts

Six (13%) of the 46 IHH patients, including five anosmic patients and one hyposmic patient, presented bilateral olfactory tract agenesis. In all cases, bilateral agenesis of olfactory tracts was accompanied by bilateral OB agenesis and bilaterally hypoplastic OS (Tables 2 and 5). The remaining 40 (87%) patients had olfactory tracts with no anomalies.

### Olfactory sulci

The average OB and pituitary volumes and OS lengths and depths according to olfactory function are shown in Table 6. The OS was bilaterally hypoplastic in 43 (93.5%) IHH patients. Aplastic sulci (i.e., with immeasurable depth or length) were not found. IHH patients had right sulci measuring 6–29 mm (mean (SD) = 18.3 (5) mm) and left sulci measuring 11–25 mm (mean (SD) = 17.4 (4) mm) (Tables 2 and 6). OS length was reduced at least unilaterally in all patients, both KS and nIHH. IHH patients had OS depths of 4–16 mm (mean (SD) – right: 9.3 (3) mm; left: 8.4 (3) mm) (Table 6). Hypoplastic sulci were detected in all 29 KS patients in both right and left hemispheres (Tables 2 and 6). Thirty-seven (80.4%) of all 46 IHH patients with OS hypoplasia had uni- or bilateral OB hypoplasia/aplasia. All subjects with abnormal OBs had OS abnormalities.

**Table 3** Genetic variants in analyzed normosmic patients with IHH.

| Patient's ID | HGVS variant | ACMG (class, evidence) | Frequency (Gnom AD) |
|---|---|---|---|
| P1 | *FGFR1 p.K256E (c.766A>G)* | VUS (PM1, PM2, PP2) | Novel |
| P2 | *WDR11 p.P3R (c.8C>G)* | VUS (PM2, PP3) | Novel |
| P3 | *NR0B1 p.N440I (c.1319A>T)* | LP (PS4, PM2, PP3) | Novel |
| P4 | *KISS1R (c.369+27G>A)* | VUS (PM2, PP3, BP7) | Novel |
| P5 | *FGFR1 p.I331T (c.992T>C)* | LP (PM1, PM2, PP2, PP3) | Novel |
| P6 | *OTX1 p.G19V (c.56G>T)* | VUS (PM2, PP3) | Novel |
| P7 | *KISS1R, (c.369+27G>A)* | VUS (PM2, PP3, BP7) | Novel |
| P8 | *TACR3: p. I249V (c.745A>G)* | LB (PS4, PM2, PP3) | Novel |
| P9 | *LHX4 p.R208 = (c.622A>C)* | VUS (PM2, PP3, BP7) | 0.000065 |
| P10 | *FGF17 (c.-38G>A)* | VUS (PM2, PP3, BP7) | 0.009 |
| P11 | *GNRHR p.R262Q. (c.416G>A)* | P (PS3, PM2, PM3, PM5, PP2, PP3) | 0.00015 |
| P12 | *TACR3 p.A36Qfs*81 (c.106delG)* | LP (PVS1, PM2) | Novel |
| P13 | *LEPR p.V754M (c.2260G>A)* | VUS (PM2, PP2, PP3) | 0.001039 |
| P14 | *SOX3 p.R155Afs*26 (c.462delG)* | P (PVS1, PM2) | Novel |
| P15 | *NSMF p.H303 = (c.909C>T)* | B (BP, BP7) | 0.00026 |
| P16 | *HS6ST1 p. R375C (c.1123C>T)* | VUS (PM2, PP2, PP3) | 0.00005511 |
| P17 | *BMP4 p.G249Afs*36 (c.746delG)* | P (PVS1, PM2) | Novel |

Table contains the most relevant genetic variants that were detected in patients. B, benign; LB, likely benign; LP, likely pathogenic; VUS, variant of uncertain significance; and P, pathogenic.

Right and left OS depths were compared to identify hemispheric asymmetry. The mean OS depth was higher in the right hemisphere in patients ($P$ = 0.03) and left hemisphere in controls (insignificant, $P$ = 0.10) (Wilcoxon's matched-pair T).

## Olfactory region cortical thickness

MRI measurements of the olfactory region cortical thickness showed a thinner cortex in IHH patients than in controls. IHH patients had thinner olfactory cortices than controls (mean (SD): 3.7 (1) mm vs 4.7 (1) mm; $P$ < 0.001).

**Table 4** Genetic variants in analyzed hyposmic/anosmic patients with IHH.

| Patient's ID | HGVS variant | ACMG (class, evidence) | Frequency (Gnom AD) | Hyposmia/anosmia |
|---|---|---|---|---|
| P18 | *WDR11 p.M769V (c.2305A>G)* | VUS (PP2, PP3, BS1) | 0.001092 | Hyposmia |
| P19 | *CHD7 p.E1195A (c.3584A>C)* | LP (PM1, PM2, PM5, PP2, PP3) | Novel | Hyposmia |
| P20 | *CHD7 p.N1030H (c.3088A>C)* | P (PS4, PM1, PM2, PM5, PP2, PP3) | Novel | Anosmia |
| P21 | *KISS1R (c.369+27G>A)* | VUS (PM2, PP3, BP7) | Novel | Anosmia |
| P23 | *CHD7 p.N1030H (c.3088A>C)* | P (PS4, PM1, PM2, PM5, PP2, PP3) | Novel | Hyposmia |
| P24 | *LHX4 p.D128 = (c.384C>T)* | B (PM2, PP3, BP7) | 0.009952 | Anosmia |
| P25 | *WDR11 p.M769V (c.2305A>G)* | VUS (PM2, PP3) | 0.001092 | Anosmia |
| P26 | *SOX3: p381 = (c.1143G>A)* | VUS (PM2, PP3) | Novel | Anosmia |
| P27 | *GLI2 p.L1488F (c.4464G>T)* | VUS (PM2, PP3) | Novel | Hyposmia |
| P28 | *IGSF10 p.A253T (c.5948T>A)* | VUS (PM2, PP3) | Novel | Hyposmia |
| P29 | *HS6ST1 p.R249S (c.745C>A)* | VUS (PM2, PP3) | Novel | Anosmia |
| P30 | *GNRHR p.R139H (c.416G>A)* | P (PS3, PM1, PM3, PM5, PP2, PP3) | 0.0002020 | Hyposmia |
| P31 | *HS6ST1 p.R249S (c.745C>A)* | VUS (PM2, PP3) | Novel | Anosmia |
| P34 | *GNRH1 p.C21Lfs*23 (c.60_61insC)* | P (PVS1, PS3, PM1, PM2, PM3, PM5, PP2, PP3) | Novel | Anosmia |
| P36 | *FGF8 intronic (c.445-62G>T)* | VUS (PM2, PP3, BP7) | Novel | Anosmia |
| P37 | *PROKR2 p.R85H (c.254G>A)* | LP (PS3, PS4, PM1, PM2, PM3, PM5) | 0.001246 | Anosmia |
| P38 | *LRRIQ3 p.R227C (c.679C>T)* | VUS (PM2, PP3) | Novel | Hyposmia |
| P40 | *FGFR1 p. R281W (c.841C>T)* | P (PM1, PM2, PM5, PP2, PP3) | Novel | Anosmia |
| P41 | *CHD7 p.N1030H (c.3088A>C)* | P (PS4, PM1, PM2, PM5, PP2, PP3) | Novel | Anosmia |
| P42 | *CHD7 p.D2838Tfs*51 (c.8512delG)* | P (PVS1, PS4, PM1, PM2, PM5, PP2, PP3) | Novel | Hyposmia |
| P43 | *GNRHR p.P146S (c.436C>T)* | VUS (PM1, PP2, PP3) | 0.001212 | Hyposmia |
| P44 | *GNRH1 p.F65 = (c.183C>T)* | VUS (PM1, PP2, PP3) | 0.004759 | Hyposmia |
| P45 | *PROKR2 p.S130= (c.390C>T)* | VUS (PM2, PP3) | 0.0008596 | Hyposmia |

Table contains the most relevant genetic variants that were detected in patients. B, benign; LB, likely benign; LP, likely pathogenic; VUS, variant of uncertain significance; and P, pathogenic. In patients P22, P32, P33, P35, P39 and P46, no genetic defects were detected in the studied panel of genes. In the case of individuals lacking a pathogenic variant, other genes should also be tested or genomic testing should be used.

**Table 5** MRI abnormalities of olfactory structures and pituitary gland hypoplasia in olfactory categories.

| No. of patients with abnormalities of | KS with anosmia | KS with hyposmia | nIHH | All IHH patients |
|---|---|---|---|---|
| Right OB | 12/16 (75%) | 9/13 (69.2%) | 9/17 (53%) | 30/46 (65.2%) |
| Right OS length | 16/16 (100%) | 13/13 (100%) | 16/17 (94.1%) | 45/46 (97.8%) |
| Right OS depth | 13/16 (81.25%) | 11/13 (84.6%) | 16/17 (94.1%) | 40/46 (87%) |
| Left OB | 14/16 (87.5%) | 10/13 (76.9%) | 13/17 (76.5%) | 37/46 (80.4%) |
| Left OS length | 16/16 (100%) | 13/13 (100%) | 17/17 (100%) | 46/46 (100%) |
| Left OS depth | 16/16 (100%) | 11/13 (84.6%) | 17/17 (100%) | 44/46 (95.7%) |
| Olfactory cortex thickness | 9/16 (56.25%) | 6/13 (46.2%) | 4/17 (23.5%) | 19/46 (41.3%) |
| Pituitary hypoplasia | 10/16 (62.5%) | 10/13 (76.9%) | 12/17 (70.6%) | 32/46 (69.6%) |

KS, Kallmann syndrome; nIHH, normosmic isolated hypogonadotropic hypogonadism; OB, olfactory bulb; and OS, olfactory sulcus.

Piriform cortex thickness did not differ significantly between nIHH and KS patients (mean (SD): 3.9 (1) mm vs 3.6 (1) mm; $P = 0.12$) (Table 5).

### Pituitary gland

IHH patients had significantly smaller pituitary volumes than controls (mean (SD): 228 (121) $mm^3$ vs 434 (88) $mm^3$; $P < 0.001$). APH (<5 mm height) was found in 30 (65.2%) IHH patients, with 15 having heights <4 mm. A decreased pituitary volume was found in 32 (69.6%) IHH patients (Table 5). No empty sella syndrome was observed.

Pineal cysts, ranging in size from 3 to 10 mm, were observed in 19.6% of IHH patients (9 out of 46: 6 in KS and 3 in nIHH). Three IHH patients had other hypothalamic–pituitary abnormalities: a non-functioning pituitary microadenoma (3 mm) in a normosmic male, ectopic posterior pituitary in an anosmic KS male and a thickened pituitary stalk in a normosmic male (Table 2).

### ELO and MRI results

All 29 dysosmic patients presented with at least one olfactory system alteration on MRI (Table 5). Of 16 anosmic patients, 15 had abnormal MRI, with OB hypoplasia or aplasia and hypoplastic sulci. Of 17 subjects, previously normosmic in ELO, 13 presented OB anomalies on MRI. MRI measurements of olfactory structures and ELO did not align in IHH, with a kappa index of −0.08 ($P > 0.05$, 95% CI: −0.23 to 0.07).

OB volume, OS length and depth, olfactory cortex thickness and pituitary height and volume showed positive correlation. Pituitary volume also correlated with OB volume, OS dimensions and olfactory cortex thickness.

## Discussion

Olfactory system MRI is useful in KS diagnostics and management (9, 30, 31). This study aimed to assess olfactory system and pituitary abnormalities on MRI in IHH patients, both normosmic and dysosmic. Age- and sex-matched control group enrollment is one of the strengths of this study.

A notable finding is almost 33% diagnostic efficiency in identifying P/LP defects in IHH. The significant incidence of VUS (nearly 50%) in IHH necessitates further investigation and observation. Gene defects corresponded to the patients' olfactory phenotype and were found not only in KS patients with distinct reproductive phenotype. The targeted gene panel was

**Table 6** MRI measurements of olfactory bulb volume, olfactory sulcus length and depth, olfactory cortex thickness and pituitary gland volume in IHH patients and controls.

| Parameter | nIHH | KS patients with hyposmia/anosmia | Controls | *P* value (nIHH vs KS) | *P* value (nIHH vs controls) | *P* value (KS vs controls) |
|---|---|---|---|---|---|---|
| Age | 28.9 (8) | 29.4 (9) | 29 (10) | 0.9 | 0.87 | 0.86 |
| Right OBV ($mm^3$) | 50.1 (18) | 31.5 (26) | 72.6 (9) | 0.01 | <0.001 | <0.001 |
| Right OS length (mm) | 20.3 (4) | 17.2 (5) | 36.9 (5) | 0.03 | <0.001 | <0.001 |
| Right OS depth (mm) | 10.5 (2) | 8.7 (3) | 15.5 (1) | 0.03 | <0.001 | <0.001 |
| Left OBV ($mm^3$) | 42.6 (17) | 25.4 (28) | 72.4 (8) | 0.01 | <0.001 | <0.001 |
| Left OS length (mm) | 18.9 (3) | 16.6 (4) | 36.5 (5) | 0.05 | <0.001 | <0.001 |
| Left OS depth (mm) | 9.4 (2) | 7.9 (3) | 15.9 (1) | 0.02 | <0.001 | <0.001 |
| Olfactory cortex thickness (mm) | 3.9 (1) | 3.6 (1) | 4.7 (1) | 0.12 | <0.001 | <0.001 |
| PGV ($mm^3$) | 243 (110) | 220 (129) | 395.7 (70) | 0.24 | <0.001 | <0.001 |

OBV, olfactory bulb volume; OS, olfactory sulcus; and PGV, pituitary gland volume.

a study limitation, and as indicated by the current research, this approach also has limited efficacy and cannot address the genetic basis for many patients with IHH (32, 33). Whole genome sequencing may be necessary in next studies.

Quantitative MRI findings showed that nearly 59% of IHH patients, including seven normosmic subjects, had bilaterally hypoplastic OBs. Buschhuter and coworkers found that in healthy individuals aged 19–79, males had average OB volumes of 70 $mm^3$ (left) and 69 $mm^3$ (right), while females had average OB volumes of 64 $mm^3$ (left) and 65 $mm^3$ (right). Significant individual differences were observed, ranging from 41 to 97 $mm^3$ (right) and from 37 to 98 $mm^3$ (left) (34). The average OB volumes in all study groups were significantly lower bilaterally compared to controls ($P < 0.010$) and were different between the nIHH and KS groups ($P = 0.01$). This is consistent with the findings by Salihoglu and coworkers, who described bilaterally lower OB volumes in nIHH and KS compared to controls (35). In that study, nIHH subjects had mean (SD) OB volumes of 56.64 (8.91) ml (right) and 56.07 (9.15) ml (left). However, the differences in right OB volumes between nIHH and control subjects were not significant (35).

According to MRI, 15 (51.7%) KS patients had bilaterally or unilaterally aplastic OBs. OB aplasia was found only in cases of concomitant dysosmia in our study. Hypoplastic and aplastic OBs and sulci have been frequently reported in KS (8, 9, 36). The prevalence of OB aplasia in this study is lower than reported, with some studies noting prevalence up to 79–87.5% in KS patients (28, 37). The lower prevalence could result from the relatively small KS groups being enrolled.

Bilateral absence of olfactory tracts was found only in KS subjects in the current study. Previous research suggests that the prevalence of olfactory tract agenesis in KS may be up to 84–90.6% (28, 37). In the current study, the prevalence of bilateral tract agenesis was lower, 21% in KS. Olfactory apparatus abnormalities in nIHH have been previously described in the literature (38, 39, 40). In contrast to previous studies, we detected no aplastic or immeasurable olfactory sulci (30). Abnormal, hypoplastic OSs were observed in all IHH patients, with 93.5% showing bilateral hypoplasia. On MRI, all OB abnormalities were associated with OS hypoplasia, and about 80% of patients with hypoplastic sulci had unilaterally or bilaterally abnormal OBs. The diminished size of the OS and OB was observed in 37.5% of nIHH patients in a study by Jagtap and coworkers (38).

Olfactory region cortical thickness was previously described in a healthy population (29). In this study, measurements of piriform cortical thickness were lower in all IHH patients than in controls ($P < 0.001$). However, piriform cortical thickness was not a useful measurement to distinguish between anosmic and hyposmic patients. A positive correlation was observed between the OB volume and piriform cortex thickness (for right OB volume: $r = 0.456$, $P = 0.002$; for left OB volume: $r = 0.418$, $P = 0.004$). On the contrary, an inverse association between the OB volume and the overlying cortical thickness was observed by Ottaviano and coworkers (41). A positive correlation between the left and right OB volume and OS length and depth, olfactory region cortical thickness and pituitary gland height observed in the current study suggests the role of the olfactory system in forebrain embryogenesis and the possible relationship between pituitary development and the formation of the olfactory apparatus.

IHH patients were classified into olfactory categories using ELO based on the availability of this olfactory test. 63.1% of patients showed abnormal olfactory function in ELO; 16 were considered anosmic and 13 were considered hyposmic. The ELO results were consistent with patients' self-reported problems with their sense of smell. However, the consistency of olfactory region MRI measurements and ELO was poor in the case of nIHH in our study. Most nIHH patients had at least one structural alteration of the olfactory system in MRI, most often affecting the OS length. The results of both subjective and objective smell tests seem to be comparable. Nevertheless, it should be remembered that the sensitivity of subjective tests could be diminished due to patients' potential manipulation or a lack of patient cooperation (42). Moderate consistency between the results of MRI quantification and the 40-item University of Pennsylvania Smell Identification Test (UPSIT) in a group of 21 KS patients and 16 healthy volunteers was described by Koenigkam-Santos and coworkers (30). A correlation between the olfactory phenotype and MRI assessments of the olfactory system in IHH was also noted by Jagtap and coworkers (38). However, normal anatomy of OBs and OSs in MRI may be observed in KS patients (30, 43). High MRI accuracy in KS diagnostics has been stressed in the literature – brain MRI is characterized by sensitivity in the range of 76–100% in KS (9, 30, 31).

Currently, there are little data on high-resolution MRI of the olfactory system in nIHH, one of the few examples being a study by Tang and coworkers, who described five female patients with nIHH with normal olfactory MRI findings (37). As mentioned earlier, bilaterally lower OB volumes in nIHH vs controls were described by Salihoglu and coworkers (35). Della Valle and coworkers found hypoplasia or agenesis of OBs in 7.6% of nIHH patients (39). OB and/or olfactory tract dysplasia was detected using MRI in a group of 11 nIHH subjects in a study by Wang and coworkers. However, in the same study, other patients presented with no OB anomalies and/or olfactory tract on MRI but reported a reduced sense of smell (28). In the current study, the prevalence of olfactory system abnormalities in nIHH is higher than previously reported, although MRI could not reliably differentiate between KS and nIHH. Weiss and coworkers suggest that the absence of apparent OBs does not always correlate

with anosmia. In fact, about 0.6% women with no OBs visualized on brain MRI were normosmic. This phenomenon was previously reported in over 4% of women but not in men (44). Due to the complexity and plasticity of the olfactory system, more studies are necessary to establish the prevalence of olfactory region abnormalities in nIHH. A shared pathophysiological origin of particular olfactory types of IHH is proposed. Hyposmic IHH subjects harbor defects in genes controlling GnRH neuronal migration and GnRH secretion. These same genes underlie the background of both nIHH and KS (45, 46, 47). A morphological assessment of the olfactory system seems insufficient, and the functional MRI should be considered in both nIHH and KS patients to measure brain activation in response to olfactory stimuli.

Several case reports were previously published on pituitary abnormalities in IHH. In the current study, we frequently observed pituitary hypoplasia in IHH, ranging from 65.2 to 69.6% (anterior pituitary height vs volume measurements). These patients might be at risk of developing combined anterior pituitary hormone deficiency, as IHH and pituitary hypoplasia may precede the development of this disorder. The prevalence of APH in our study is higher than previously reported. Tang and coworkers reported APH in 12.5% of female Chinese IHH patients who underwent hypothalamic–pituitary region MRI (37). Zhang and coworkers described an incidence of APH in 37.5% of KS subjects (11). Diminished stimulation due to the lack of GnRH and hypothalamic GnRH neurons could be a reason for pituitary hypoplasia in IHH (48). Moreover, there was no significant correlation between the incidence of pituitary hypoplasia and the olfactory category of IHH subjects in the current study. Although empty sella syndrome was previously described in up to 7.1% of IHH patients, empty sella syndrome was not observed in the current study (24).

Other pituitary abnormalities were described in just 6.5% of the enrolled IHH patients, including one case of an ectopic posterior pituitary, one non-functioning microadenoma and one thickened pituitary stalk. Both the ectopic posterior pituitary gland and thickened pituitary stalks are rare findings (49, 50, 51). The incidence of microadenoma in our study is low. In comparison, Bolu and coworkers reported microadenomas and an irregularly contrasting pituitary in 18.2 and 10.7% of IHH patients. Meanwhile, Bonomi and coworkers observed non-functional microadenomas in 5–14% of IHH cases (13, 52). Pineal cysts were identified in 6 KS and 3 nIHH subjects, with a reported incidence in healthy adults ranging from 0.58 to 23%, showing higher prevalence in females and older patients (53, 54, 55).

The study limitations include patient recruitment and IHH diagnostic criteria. In our study, nearly 30% had adult-onset IHH. Adult-onset IHH is typically rare, and it might be overdiagnosed in our study (56). Furthermore, only the ELO test was used for smell identification, with no other validation methods (such as the University of Pennsylvania Smell Identification Test) included. ELO has limited sensitivity, which can explain the differences between its results and MRI findings. Finally, the use of a ready-made NGS panel may be a limiting factor due to the lack of coverage for genes very rarely associated with IHH.

## Conclusions

MRI constitutes a safe and cost-effective method for morphological analysis of the pituitary and olfactory system. IHH has characteristic features on MRI, including APH as part of its clinical picture. OB abnormalities are surprisingly frequent in IHH, occurring more commonly in KS compared to nIHH. This observation emphasizes the importance of pituitary and olfactory region visualization in both dysosmic and normosmic IHH subjects, although olfactory system abnormalities can be found in both conditions and the differentiation of KS and nIHH is not based solely on MRI findings. MRI should be considered a method of choice in IHH diagnostics, as it is highly useful for assessing patients with hypoplastic pituitary and olfactory region disorders. The MRI phenotypic spectrum of the olfactory system and pituitary gland may guide additional diagnostics and genetic testing in IHH. Further studies on MRI phenotypes in IHH are needed.

**Supplementary materials**

This is linked to the online version of the paper at https://doi.org/10.1530/EC-24-0437.

**Declaration of interest**

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the work.

**Funding**

The APC was funded by Sanofi.

**Author contribution statement**

Conceptualization was done by MK and KZ. MK, KK, BB, JM and KZ designed the methodology. Formal analysis and investigation was performed by MK, KK, JM, MRa, PKK and JK. Data curation was performed by MK and KK. BB helped with software. MK, BB and PK wrote the original draft. MK, BB, PK, PKK, KK, KZ, JM, MRa and MRu reviewed and edited the manuscript. KZ and MRu supervised the study. MK was involved in project administration.

**Data availability**

The datasets generated during and/or analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

**Institutional review board statement**

The consent of the Bioethical Committee at Poznan University of Medical Sciences was granted by the decision of the resolution no. 1002/13 of 05 December 2013 and no. 990/15 of 05 November 2015. The study was conducted in accordance with the Declaration of Helsinki.

**Informed consent statement**

Informed consent was obtained from all subjects involved in the study.

**Acknowledgments**

The authors would like to thank Mark Jensen for professional language verification of the article.

## References

1 Balasubramanian RCW & Crowley WF Jr. Isolated gonadotropin-releasing hormone (GnRH) deficiency. In *GeneReviews*. Seattle, WA, USA: University of Washington. (https://www.ncbi.nlm.nih.gov/books/NBK1334/)

2 Bianco SD & Kaiser UB. The genetic and molecular basis of idiopathic hypogonadotropic hypogonadism. *Nat Rev Endocrinol* 2009 **5** 569–576. (https://doi.org/10.1038/nrendo.2009.177)

3 Fromantin M, Gineste J, Didier A, *et al.* Impuberism and hypogonadism at induction into military service. Statistical study. *Probl Actuels Endocrinol Nutr* 1973 **16** 179–199.

4 Laitinen EM, Vaaralahti K, Tommiska J, *et al.* Incidence, phenotypic features and molecular genetics of Kallmann syndrome in Finland. *Orphanet J Rare Dis* 2011 **6** 41. (https://doi.org/10.1186/1750-1172-6-41)

5 Nair S, Jadhav S, Lila A, *et al.* Spectrum of phenotype and genotype of congenital isolated hypogonadotropic hypogonadism in Asian Indians. *Clin Endocrinol* 2016 **85** 100–109. (https://doi.org/10.1111/cen.13009)

6 Suzuki M, Takashima T, Kadoya M, *et al.* Height of normal pituitary gland on MR imaging: age and sex differentiation. *J Comput Assist Tomogr* 1990 **14** 36–39. (https://doi.org/10.1097/00004728-199001000-00006)

7 Suzuki M, Takashima T, Kadoya M, *et al.* MR imaging of olfactory bulbs and tracts. *AJNR Am J Neuroradiol* 1989 **10** 955–957.

8 de m Freitas P, Carvalho S, Ribeiro F, *et al.* Neuroradiology of Kallmann's syndrome. *Acta Med Port* 2001 **14** 123–126.

9 Truwit CL, Barkovich AJ, Grumbach MM, *et al.* MR imaging of Kallmann syndrome, a genetic disorder of neuronal migration affecting the olfactory and genital systems. *AJNR Am J Neuroradiol* 1993 **14** 827–838.

10 Ach T, Marmouch H, Elguiche D, *et al.* A case of Kallmann syndrome associated with a non-functional pituitary microadenoma. *Endocrinol Diabetes Metab Case Rep* 2018 **2018** 18-0027. (https://doi.org/10.1530/edm-18-0027)

11 Zhang Z, Sun X, Wang C, *et al.* Magnetic resonance imaging findings in Kallmann syndrome: 14 cases and review of the literature. *J Comput Assist Tomogr* 2016 **40** 39–42. (https://doi.org/10.1097/rct.0000000000000334)

12 Dallago CM, Abech DD, Pereira-Lima JF, *et al.* Two cases of Kallmann syndrome associated with empty sella. *Pituitary* 2008 **11** 109–112. (https://doi.org/10.1007/s11102-007-0043-9)

13 Bolu SE, Tasar M, Uckaya G, *et al.* Increased abnormal pituitary findings on magnetic resonance in patients with male idiopathic hypogonadotrophic hypogonadism. *J Endocrinol Invest* 2004 **27** 1029–1033. (https://doi.org/10.1007/bf03345305)

14 Jonklaas J. Atypical presentation of a patient with both Kallmann syndrome and a craniopharyngioma: case report and literature review. *Endocr Pract* 2005 **11** 30–36. (https://doi.org/10.4158/ep.11.1.30)

15 Balasubramanian R & Crowley WF Jr. Isolated GnRH deficiency: a disease model serving as a unique prism into the systems biology of the GnRH neuronal network. *Mol Cell Endocrinol* 2011 **346** 4–12. (https://doi.org/10.1016/j.mce.2011.07.012)

16 Lin J, Mao J, Wang X, *et al.* Optimal treatment for spermatogenesis in male patients with hypogonadotropic hypogonadism. *Medicine* 2019 **98** e16616. (https://doi.org/10.1097/md.0000000000016616)

17 Silveira LFG & Latronico AC. Approach to the patient with hypogonadotropic hypogonadism. *J Clin Endocrinol and Metab* 2013 **98** 1781–1788. (https://doi.org/10.1210/jc.2012-3550)

18 Raivio T, Falardeau J, Dwyer A, *et al.* Reversal of idiopathic hypogonadotropic hypogonadism. *N Engl J Med* 2007 **357** 863–873. (https://doi.org/10.1056/nejmoa066494)

19 Kaluzna M, Budny B, Rabijewski M, *et al.* Defects in GnRH neuron migration/development and hypothalamic-pituitary signaling impact clinical variability of kallmann syndrome. *Genes* 2021 **12** 868. (https://doi.org/10.3390/genes12060868)

20 Kałużna M, Budny B, Rabijewski M, *et al.* Variety of genetic defects in GnRH and hypothalamic-pituitary signaling and development in normosmic patients with IHH. *Front Endocrinol* 2024 **15** 1396805. (https://doi.org/10.3389/fendo.2024.1396805)

21 Kałużna M, Budny B, Rabijewski M, *et al*. Variety of genetic defects in GnRH and hypothalamic-pituitary signaling and development in normosmic patients with IHH. *Front Genet* 2024 **15** 1396805. (https://doi.org/10.3389/fendo.2024.1396805)

22 Elsberg CA, Levy I & Brewer ED. A new method for testing the sense of smell and for the establishment of olfactory values of odorous substances. *Science* 1936 **83** 211–212. (https://doi.org/10.1126/science.83.2148.211)

23 Obrebowski A, Pruszewicz A, Flieger S, *et al.* Value of olfactometry and gustometry in the evaluation of delayed complications following facial trauma. *Czas Stomatol* 1978 **31** 1047–1049.

24 Pruszewicz A. Apropos of gustatory and olfactory tests. *Otolaryngol Pol* 1965 **19** 29–37.

25 Obrębowski A, Świdziński T & Świdziński P. Przydatność kliniczna olfaktometrii elektrofizjologicznej. *Postępy W Chirurgii Głowy I Szyi* 2014 **13** 15–21.

26 Murray RA, Maheshwari HG, Russell EJ, *et al.* Pituitary hypoplasia in patients with a mutation in the growth hormone-releasing hormone receptor gene. *AJNR Am J Neuroradiol* 2000 **21** 685–689.

27 Rombaux P, Duprez T & Hummel T. Olfactory bulb volume in the clinical assessment of olfactory dysfunction. *Rhinology* 2009 **47** 3–9.

28 Hacquart T, Ltaief-Boudrigua A, Jeannerod C, *et al.* Reconsidering olfactory bulb magnetic resonance patterns in Kallmann syndrome. *Ann Endocrinol* 2017 **78** 455–461. (https://doi.org/10.1016/j.ando.2016.12.003)

29 Olofsson JK, Rogalski E, Harrison T, *et al.* A cortical pathway to olfactory naming: evidence from primary progressive aphasia. *Brain* 2013 **136** 1245–1259. (https://doi.org/10.1093/brain/awt019)

30 Koenigkam-Santos M, Santos AC, Versiani BR, *et al.* Quantitative magnetic resonance imaging evaluation of the olfactory system in Kallmann syndrome: correlation with a clinical smell test. *Neuroendocrinology* 2011 **94** 209–217. (https://doi.org/10.1159/000328437)

31 Knorr JR, Ragland RL, Brown RS, *et al.* Kallmann syndrome: MR findings. *AJNR Am J Neuroradiol* 1993 **14** 845–851.

32 Xie YD, Zheng RZ, Han HJ, *et al.* Analysis of PROKR2 gene mutation in patients with hypogonadotropic hypogonadism. *Zhonghua Nei Ke Za Zhi* 2022 **61** 933–936. (https://doi.org/10.3760/cma.j.cn112138-20210821-00571)

33 Cho CY, Tsai WY, Lee CT, *et al.* Clinical and molecular features of idiopathic hypogonadotropic hypogonadism in Taiwan: a single center experience. *J Formos Med Assoc* 2022 **121** 218–226. (https://doi.org/10.1016/j.jfma.2021.03.010)

34 Buschhuter D, Smitka M, Puschmann S, *et al.* Correlation between olfactory bulb volume and olfactory function. *Neuroimage* 2008 **42** 498–502. (https://doi.org/10.1016/j.neuroimage.2008.05.004)

35 Salihoglu M, Kurt O, Ay SA, *et al.* Retro- and orthonasal olfactory function in relation to olfactory bulb volume in patients with hypogonadotrophic hypogonadism. *Braz J Otorhinolaryngol* 2018 **84** 630–637. (https://doi.org/10.1016/j.bjorl.2017.07.009)

36 Stern Y, Egelhoff J & Shott SR. Imaging quiz case 1. Absence of the olfactory bulb and tracts consistent with Kallmann syndrome. *Arch Otolaryngol Head Neck Surg* 1998 **124** 342–343.

37 Tang RY, Chen R, Ma M, *et al.* Clinical characteristics of 138 Chinese female patients with idiopathic hypogonadotropic hypogonadism. *Endocr Connect* 2017 **6** 800–810. (https://doi.org/10.1530/ec-17-0251)

38 Jagtap VS, Sarathi V, Lila AR, *et al.* An objective olfactory evaluation and its correlation with magnetic resonance imaging findings in Asian Indian patients with idiopathic hypogonadotropic hypogonadism. *Endocr Pract* 2013 **19** 669–674. (https://doi.org/10.4158/ep13008.or)

39 Della Valle E, Vezzani S, Rochira V, *et al.* Prevalence of olfactory and other developmental anomalies in patients with central hypogonadotropic hypogonadism. *Front Endocrinol* 2013 **4** 70. (https://doi.org/10.3389/fendo.2013.00070)

40 Wang Y, Gong C, Qin M, *et al.* Clinical and genetic features of 64 young male paediatric patients with congenital hypogonadotropic hypogonadism. *Clin Endocrinol* 2017 **87** 757–766. (https://doi.org/10.1111/cen.13451)

41 Ottaviano G, Cantone E, D'Errico A, *et al.* Sniffin' sticks and olfactory system imaging in patients with Kallmann syndrome. *Int Forum Allergy Rhinol* 2015 **5** 855–861. (https://doi.org/10.1002/alr.21550)

42 Anik A, Catli G, Abaci A, *et al.* Olfactory dysfunction in children with Kallmann syndrome: relation of smell tests with brain magnetic resonance imaging. *Hormones* 2015 **14** 293–299. (https://doi.org/10.14310/horm.2002.1562)

43 Ghadami M, Majidzadeh AK, Morovvati S, *et al.* Isolated congenital anosmia with morphologically normal olfactory bulb in two Iranian families: a new clinical entity? *Am J Med Genet A* 2004 **127A** 307–309. (https://doi.org/10.1002/ajmg.a.30025)

44 Weiss T, Soroka T, Gorodisky L, *et al.* Human olfaction without apparent olfactory bulbs. *Neuron* 2020 **105** 35–45.e5. (https://doi.org/10.1016/j.neuron.2019.10.006)

45 Maione L, Dwyer AA, Francou B, *et al.* Genetics in endocrinology: genetic counseling for congenital hypogonadotropic hypogonadism and Kallmann syndrome: new challenges in the era of oligogenism and next-generation sequencing. *Eur J Endocrinol* 2018 **178** R55–R80. (https://doi.org/10.1530/eje-17-0749)

46 Kim JH, Seo GH, Kim GH, *et al.* Targeted gene panel sequencing for molecular diagnosis of Kallmann syndrome and normosmic idiopathic hypogonadotropic hypogonadism. *Exp Clin Endocrinol Diabetes* 2018 **127** 538–544. (https://doi.org/10.1055/a-0681-6608)

47 Topaloglu AK. Update on the genetics of idiopathic hypogonadotropic hypogonadism. *J Clin Res Pediatr Endocrinol* 2018 **9** 113–122. (https://doi.org/10.4274/jcrpe.2017.s010)

48 Tsutsumi R & Webster NJ. GnRH pulsatility, the pituitary response and reproductive dysfunction. *Endocr J* 2009 **56** 729–737. (https://doi.org/10.1507/endocrj.k09e-185)

49 Mitchell LA, Thomas PQ, Zacharin MR, *et al.* Ectopic posterior pituitary lobe and periventricular heterotopia: cerebral malformations with the same underlying mechanism? *AJNR Am J Neuroradiol* 2002 **23** 1475–1481.

50 Lahiri AK, Sundareyan R, Jenkins D, *et al.* MRI of ectopic posterior pituitary gland with dysgenesis of pituitary stalk in a patient with hypogonadotropic hypogonadism. *Radiol Case Rep* 2018 **13** 764–766. (https://doi.org/10.1016/j.radcr.2018.05.004)

51 Hirsch D, Benbassat C, Toledano Y, *et al.* Pituitary imaging findings in male patients with hypogonadotrophic hypogonadism. *Pituitary* 2015 **18** 494–499. (https://doi.org/10.1007/s11102-014-0601-x)

52 Bonomi M, Vezzoli V, Krausz C, *et al.* Characteristics of a nationwide cohort of patients presenting with isolated hypogonadotropic hypogonadism (IHH). *Eur J Endocrinol* 2018 **178** 23–32. (https://doi.org/10.1530/eje-17-0065)

53 Pu Y, Mahankali S, Hou J, *et al.* High prevalence of pineal cysts in healthy adults demonstrated by high-resolution, noncontrast brain MR imaging. *AJNR Am J Neuroradiol* 2007 **28** 1706–1709. (https://doi.org/10.3174/ajnr.a0656)

54 Al-Holou WN, Garton HJ, Muraszko KM, *et al.* Prevalence of pineal cysts in children and young adults. Clinical article. *J Neurosurg Pediatr* 2009 **4** 230–236. (https://doi.org/10.3171/2009.4.peds0951)

55 Gokce E & Beyhan M. Evaluation of pineal cysts with magnetic resonance imaging. *World J Radiol* 2018 **10** 65–77. (https://doi.org/10.4329/wjr.v10.i7.65)

56 Dwyer AA, Hayes FJ, Plummer L, *et al.* The long-term clinical follow-up and natural history of men with adult-onset idiopathic hypogonadotropic hypogonadism. *J Clin Endocrinol Metab* 2010 **95** 4235–4243. (https://doi.org/10.1210/jc.2010-0245)